A Real Dracula Story

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Research, discovery and publishing in a prestigious journal are not as easy as 1-2-3. However, an outstanding mentor and a well-equipped lab made this feat possible for doctoral student Fei Jiang.

The mentor is Myron F. Goodman, USC College professor of biological sciences and chemistry, and the lab where it all happened is in Ray R. Irani Hall on University Park Campus.

In a recent interview, Goodman and Jiang explained the intricacies of their research in a July 2009 *Nature* article, “The active form of DNA polymerase V is UmuD’ 2C-RecA-ATP.” Whew! That’s quite a mouthful for non-scientists, but hang in there.

Pol V, an enzyme discovered by Goodman’s research group in 1999, activates when a cell’s DNA is damaged. Essentially the damaged cell sends out an SOS and RecA comes to the rescue.

The enzyme RecA is a paradigm of sorts for understanding how a protein can catalyze homologous genetic recombination. And, genetic recombination is the process in which a strand of genetic material is broken and then joined to a different DNA molecule.

Pol V, an Oscar Madison of sorts in the cell world, has been coined the “sloppier copier” because it generates mistakes as it duplicates.

“Two and a half years ago I set out to prove the observation of the then doctoral student of Professor Goodman, Kathi Schlacher, of how Pol V

works when a cell is damaged from radiation or other serious blows and then kick starts the repair process." Jiang said. "Kathy realized that RecA did not need to be on the DNA itself for the repair to occur."

Jiang proved Schlacker's observations as true.

But, how did it happen?

RecA is the subject of one of the two longstanding enigmas that Goodman and Jiang sought to solve in their study. RecA always seemed to be present around DNA repair sites, but why? The second question focused on discovering the specific composition of the active form of Pol V.

"Three years ago Kathi Schlacher realized that maybe you don't need the RecA on the DNA that is being copied and that it could be somewhere else in the cell," Goodman said. If this is true, Goodman and Schlacher wondered where the single-stranded DNA comes from that the RecA is going to form to interact with the DNA?

Jiang found the mechanism — no matter where a RecA protein filament is located, whether on the same DNA being copied or remote in the cell, a single RecA molecule along with an ATP molecule gets transferred over to Pol V and converts it into a live enzyme.

"So basically it's the Dracula story," Goodman said. "RecA along with an important energy carrying molecule, ATP, transfers to this Pol V and then it activates. It then goes swimming around until it finds where it has to go and can copy the DNA."

When the Pol V has done its job, it becomes inactive until it is called upon again and gets another infusion from RecA. In other words, you can't kill Dracula.

“The devil is in the details and the devil is the following: previously the only way you could get an enzyme to copy this DNA, damaged or undamaged, is if you had this RecA bound to a single-stranded DNA,” Goodman said. “Whether activation on the same or different piece of DNA is being copied, it was always there so no one knew where it was coming from.”

To prove this, Jiang made DNA that could be bound by RecA and placed it on a resin for a sequence of minutes. “I exposed enzymes to this resin with the DNA on it and could centrifuge it down, then throw away RecA-DNA resin, and come to this conclusion.” To validate her findings, she conducted the experiment several times over.

But what of all the mutations made by the sloppier copier Pol V?

“One in a million mutations can be evolutionary,” Goodman said. “They may help the organism survive in the future. While mutations are almost always dangerous, an organism tolerates a certain amount of damage to its DNA. The replication of genome DNA allows for generation of a very few number of mutations that can provide the cell with a fitness advantage.”

During the two and a half year process, Jiang said there were both exciting and frustrating times.

“The reason I love science is discovery,” she said. “When you are right, it is exciting. And when you aren’t, the answer you found will lead to another door.”

Jiang found Goodman’s sense of humor refreshing and uplifting when she hit difficult places along the way.

“Among scientists in my field, we see three things as important: death,

taxes and ultimately there's RecA," Goodman chuckled.

Provided by USC College